

# Advance your allergy therapeutic development using our robust human FcεRI reporter platform

The FcεRI (Fc epsilon RI) – IgE axis is the central pathway underlying allergic disease. FcεRI, is the high affinity receptor for IgE on mast cells and basophils, is a clinically validated target for therapies designed to prevent or suppress allergic inflammation.

## Why Target the FcεRI – IgE Axis?

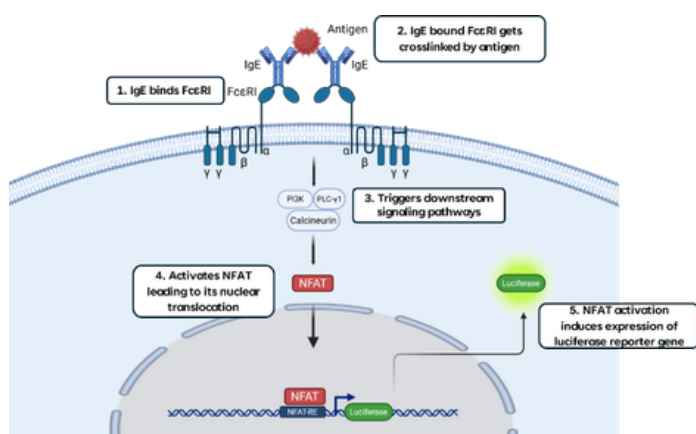
The FcεRI – IgE axis is central to allergic responses. A growing number of biologics and small molecules are designed to interrupt this pathway at different stages, prevent immune cell activation and reducing allergic responses.

| Therapeutic modality         | Mechanism of Action                                                                                 |
|------------------------------|-----------------------------------------------------------------------------------------------------|
| Anti-IgE antibodies          | Neutralise circulating IgE, preventing FcεRI sensitisation and reducing receptor expression         |
| IgE- FcεRI dissociators      | Remove pre-bound IgE from FcεRI, enabling rapid interruption of allergic responses                  |
| Downstream kinase inhibitors | Block intracellular FcεRI signalling (e.g., Syk, BTK), preventing mast cell and basophil activation |
| FcεRI-blocking Fabs          | Prevent IgE binding directly at the receptor without triggering receptor activation                 |

## Why This Matters

- FcεRI – IgE is a validated therapeutic target across multiple allergic diseases
- Diverse therapeutic modalities are being developed to interrupt this pathway through distinct mechanisms
- Functional assays that measure FcεRI – IgE activity are critical for evaluating therapeutic efficacy and safety profile, accelerating allergy drug development.

**RoukenBio has developed a high-throughput human FcεRI reporter platform by engineering Rat Basophil Leukaemic (RBL-2H3) cells to express human FcεRI and an NFAT-driven luciferase reporter, enabling sensitive and quantitative measurement of IgE-mediated receptor activation through luminescence.**

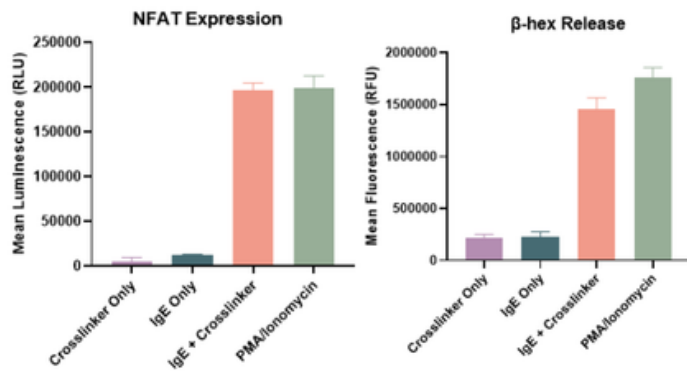


**Figure 1:** Antigen-mediated crosslinking of FcεRI-bound IgE activates NFAT signalling, inducing luciferase expression as a measure of receptor activation.

## Key Applications

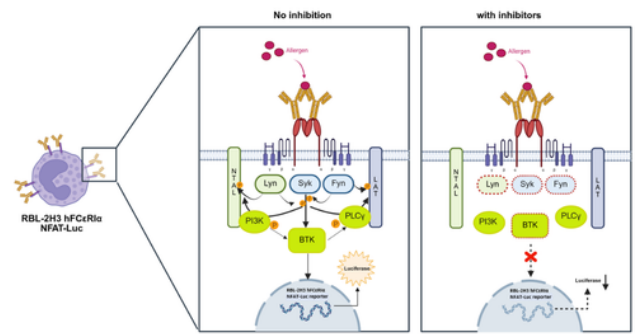
- Anti-IgE therapeutic screening
- Degranulation inhibition assessment
- Anaphylaxis liability assessment
- FcεRI signalling pathway characterisation
- Potency and mechanism-of-action studies

## NFAT signal correlates with FcεRI mediated degranulation



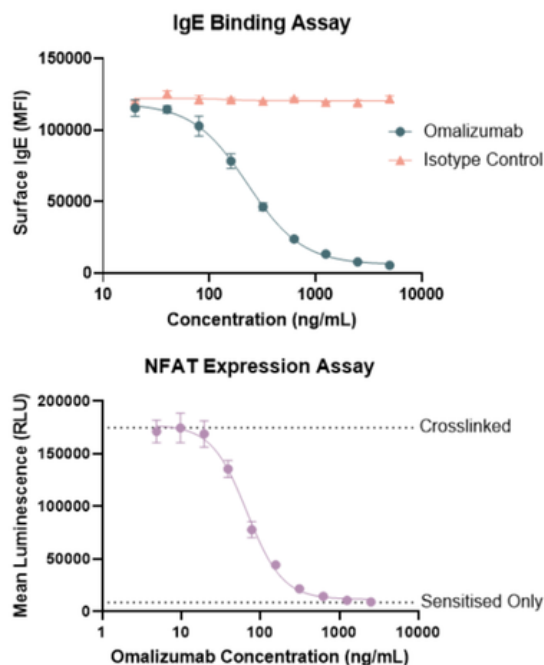
**Figure 2:** IgE-dependent receptor crosslinking drives both NFAT activation and  $\beta$ -hexosaminidase release, a widely accepted marker of degranulation, supporting the use of the reporter assay as a quantitative surrogate for allergic cell activation.

## Targeting FcεRI downstream signalling



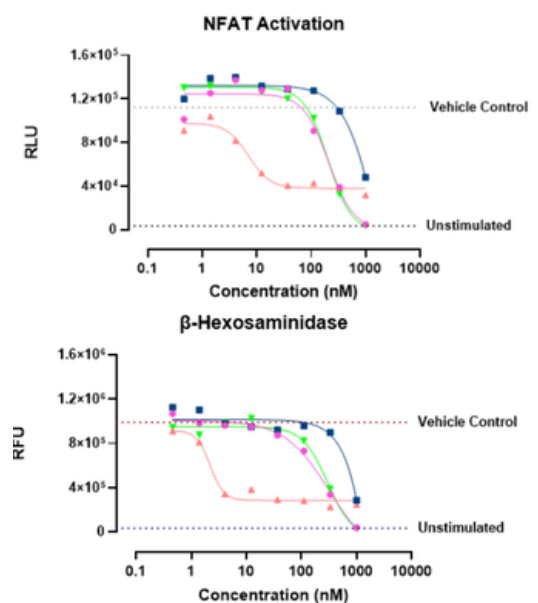
**Figure 4:** Schematic of FcεRI signalling and pathway inhibition using engineered RBL-2H3 hFcεRIa reporter cells. **Left:** Allergen-induced crosslinking of FcεRI-bound IgE activates Lyn/Fyn, Syk, PI3K, PLCγ and BTK, leading to NFAT activation and luciferase expression. **Right:** Inhibitors targeting FcεRI signalling components (e.g., Lyn, Syk or BTK) block signal transduction, reducing NFAT activation and luciferase reporter signal.

## Omalizumab inhibits IgE binding to FcεRI in a dose dependent manner



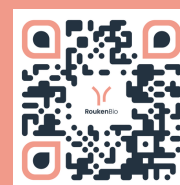
**Figure 3:** Allergen-mediated crosslinking of FcεRI receptor-bound IgE generates a strong NFAT reporter response. Treatment with omalizumab inhibits IgE binding and correspondingly suppresses NFAT activation, demonstrating a clear dose-response from the crosslinked positive control to baseline signal levels.

## Downstream Kinase inhibitors dose dependently inhibit NFAT & β-hexosaminidase



**Figure 5:** Allergen-mediated crosslinking of FcεRI receptor-bound IgE generates a strong NFAT activity and  $\beta$ -hexosaminidase release. Treatment with different TKI, Syk, BTK and IRE1a kinase inhibitors inhibits downstream signalling and correspondingly suppresses NFAT activation and  $\beta$ -hexosaminidase release, demonstrating a clear dose-response from the crosslinked positive control to baseline signal levels.

To explore how this model could de-risk and accelerate your allergy therapeutics, scan the QR code and visit our allergy page.



## RoukenBio - The CRO redefined

Backed by our brilliant minds, we have turned the traditional CRO model on its head, by fostering a collaborative and personalised approach. Our mission is simple:

**Solve problems. Deliver quality data. Propel your drug discovery breakthroughs.**

United by a passion to make sense of complexities and overcome challenges, we apply our specialised knowledge to big-picture thinking. We will explore every option to deliver over and above for your project.